



IR Micro-Spectroscopy: A Molecular Probe with Micron Resolution Workshop, May 23 and 24, 2001

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The NSLS has hosted infrared (IR) workshops during the Annual Users' Meeting for the past 4 years. This year, the workshop focused on IR *micro*-spectros-

copy, featuring scientific results from a number of disciplines along with presentations on instrumentation

improvements. Over 50 participants attended the workshop.

The scientific program began with Prof. George Flynn (SUNY at Plattsburgh). He and his colleagues at NASA perform vibrational spectroscopy on interplanetary particles. Orbiting spacecraft, as well as aircraft flying in the Earth's upper atmosphere, collect these extremely small specimens. Infrared microspectroscopy is one of several techniques that are applied to a given specimen. A particularly interesting group of particles is known as "GEMS" (glass with embedded metals and sulfides). The strong Si-O vibrational absorption for these particles matches the interplanetary dust particles. In contrast, various silicates found on Earth do not match. Research efforts are now focused on the Fe-S vibration at much lower frequencies.

Dr. Richard Newman (Museum of Fine Arts – Boston) described the use of infrared microspectroscopy for the analysis and identification of both organic and inorganic materials found in various works of art. This is particularly useful for characterizing the materials used in restoration. For some works of art, the restoration process has occurred many times over a period of centuries. In one example, Dr. Newman identified a form of calcium carbonate found in seashells used to form a white pigment. He noted that IR microspectroscopy is the most used technique in their laboratory.

Dr. Rina Dukor (Vysis, Inc.) surveyed the infrared spectra of biological systems as a lead-in to her study of cancerous tissues. IR spectra of tissues contain contributions from components such as proteins, lipids, carbohydrates and nucleic acids. Tissues are made of cells and the surrounding extracellular matrix, both of which are very inhomogeneous in composition. So, Dr. Dukor concluded that a meaningful IR study of cancerous cells must be done by probing individual cells rather than by surveying over large numbers of cells. She emphasized that this type of work can only be done utilizing a synchrotron IR source.

Prof. Max Diem (CUNY – Hunter College) gave the second talk on biological materials. Prof. Diem is also interested in the composition of tumor cells, especially the spectral signatures for nucleic acids (RNA and DNA). Researchers have been searching for differences in the nucleic acid spectra associated with various diseases, including cancer. Prof. Diem noted that the DNA contribution to an IR spectrum is often "hidden," i.e. when contained in an acquiescent nucleus, DNA is packed into separate volumes each less than 500 nm in diameter, for which the optical density would be approximately 50. Conversely, the DNA in cells that are actively replicating is not tightly packed, so the IR signatures are increased. In general, cancerous cells are rapidly dividing and IR spectral differences in tumor cells are likely a reflection of this process.

Ying Mei, a student from the laboratory of Prof. Richard Gross (Polytechnic Univ.), described his IR imaging studies on immobilized enzymes used to catalyze biopolymerization reactions. Since these reactions often occur best at high temperatures and/or in organic solvents, the enzyme is covalently bound to a polymer bead to prevent denaturation during the reaction. A typical bead is 500 microns in diameter, but IR microspectroscopy and imaging has shown that only the outer 80-100 microns actually serve to catalyze the polymerization process.

Prof. Mark Braiman (Syracuse Univ.) is developing planar infrared waveguides to study the surfaces of individual biological cells. The specimen for study is placed on the guide and sensed via attenuated total reflection (ATR) spectroscopy. Tapering the waveguide improves the angle of incidence and number of internal reflections, both of which increase the sensitivity. Prof. Braiman uses the narrow synchrotron beam to characterize the optical acceptance and throughput of these light guides.

Dr. Larry Carr (NSLS) gave the first of the instrumentation and technique talks. Larry reviewed the spatial resolution limits for IR microspectroscopy based on diffraction theory. He noted that a confocal microscope offers improvements in both resolution (narrower pattern) and contrast (significantly reduced diffraction "rings"). Efforts to improve the far-field spatial resolution by immersion in a high-index medium were also described.

Near-field approaches have the potential for surpassing the diffraction-limit by factors of 10 or more. Dr. Chris Michaels (NIST – Gaithersburg) presented results of scanning near-field microscopy using a Ti:sapphire laser and OPA system. They use infrared fibers with a taper optimized for good throughput. Variations in the refractive index, rather than the material's absorbance, produce chemical contrast in this technique. Significantly higher brightness is available from infrared free electron lasers (FELs). Dr. Ed Gillman (TJNAF) described the IR FEL at Jefferson Lab and their near-field microscope setup. They use an apertureless technique where the infrared is scattered from a small metal structure placed near the sample.

Following a lively dinner at a local Chinese restaurant, the workshop continued with 2 days of hands-on IR microspectroscopy. A number of workshop participants took advantage of this beamtime and brought samples to examine on the IR microscopes at U10B, U2A, and U2B. Particular thanks are given to Ned Marinkovic and Zhenxian Liu for giving their time and expertise at their beamlines. As organizers, we would also like to thank Simon Bare and Dan Fischer for organizing a very successful meeting this year.